Diet and Feeding of Murray Cod (*Maccullochella peelii*) Larvae

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Murray cod, *Maccullochella peelii*, larvae commence feeding on zooplankton at the completion of yolk sac absorption, 9 to 11 days after hatching at water temperatures of 20-22°C; zooplankters ranged in length from 180-450 μm. In earthen ponds, larvae fed mainly on the calanoid copepods, *Boeckella fluvialis* and *Calamoecia lucasi* and the cladocerans, *Moina micrura* and *Daphnia carinata* for two weeks; then chironomid larvae and aquatic insects became the major components of their diet. The density of zooplankton at the commencement of feeding did not affect survival in 70-L aquaria; but when initial feeding was delayed for five or more days, survival was significantly lower ($P<0.01$) at 250 zooplankters/L compared to 3000/L. A delay of 13 days resulted in high mortality irrespective of zooplankton density. These findings are briefly discussed in relation to the reduced abundance of *M. peelii* in the wild, and the development of artificial breeding techniques.

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INTRODUCTION

Little is known about the larvae of Australian native freshwater fishes, despite the importance of this stage of the life cycle. The survival of the larvae of many fish species, both in the wild and under culture conditions, is affected by the availability of food during a 'critical period' after yolk sac absorption is completed (May, 1974). Laboratory studies have shown that survival of the larvae of some species is dependent on the presence of high densities of food when exogenous feeding commences (O'Connell and Raymond, 1970; Saksena and Houde, 1972; Houde, 1975) and that a delay in the initial feeding may result in reduced survival (Houde, 1974; Laurence, 1974; Roberts *et al.*, 1978).

The successful propagation of fish requires the development of techniques for rearing the larvae. Research into the diet and factors affecting larval survival often produces biological information which, besides enabling the rearing of larvae, provides a better understanding of the life cycle and population dynamics of the species.

The Murray cod, *Maccullochella peelii* (Mitchell, 1838) is Australia's largest and most famous inland fish; it is keenly sought by commercial and recreational fishermen. There has been a dramatic decline in the abundance of Murray cod during the 1900's (Rowland, 1989a) and despite the importance of this species, little is known of its life history. There is no record of Murray cod eggs or larvae being sampled from the wild. Techniques have now been developed for the artificial spawning of *M. peelii* (Cadwallader and Gooley, 1985; Rowland, 1985, 1988). As part of studies on the biology and artificial breeding of *M. peelii*, experiments were conducted to determine the diet of larvae and effects of food density and delayed feeding on survival. The results of this research are presented and discussed below.

MATERIALS AND METHODS

Experiments were conducted in the laboratory and earthen ponds at the Inland Fisheries Research Station, Narrandera, N.S.W., using larvae from both hormone-induced breeding trials and natural spawnings in ponds (Rowland, 1983, 1988). In all

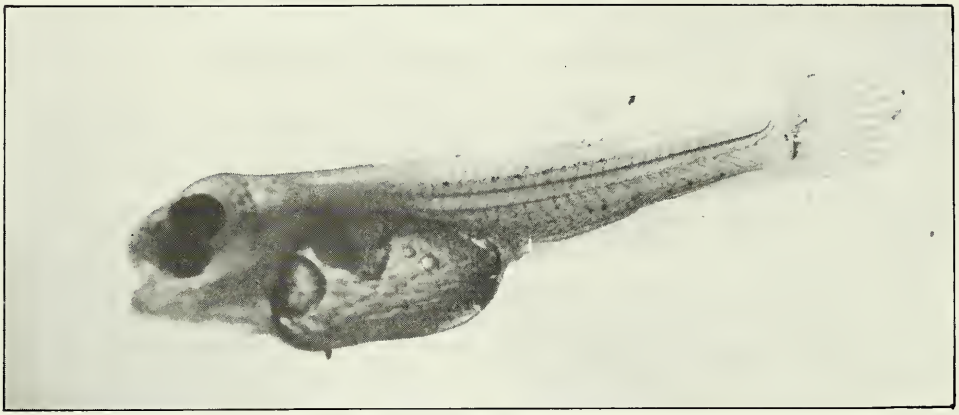

Fig. 1. Murray cod larva prior to feeding — note the large yolk sac still present.

experiments, day 1 refers to the tenth day after the completion of hatching. Water from the Murrumbidgee River was used in all rearing facilities.

Laboratory Experiments

The 70-L rectangular, glass aquaria used in feeding experiments contained sub-gravel filtration and two airstones to provide gentle circulation and aeration. The back and sides of each aquarium were covered with black polythene sheeting. The water temperature was maintained at 20-22°C. Aquaria were subject to natural photoperiod with additional overhead fluorescent lighting for 8-10 h during daylight hours. Dead plankton and debris were siphoned from each aquarium daily, and larvae and fry were given a prophylactic NaCl bath (5 g/L) for 1 h every five days to prevent infestations of protozoan parasites (Rowland and Ingram, 1991).

Larvae in aquaria were fed wild zooplankton collected from ponds on the research station using a 100 μm-mesh plankton net. The plankton was washed through Endecott sieves of mesh sizes 1000 and 500 μm. For the first two days larvae were offered only zooplankton that passed through the 500 μm sieve; from the third day all sizes of zooplankton were offered to the larvae. The density of zooplankton in each aquarium was estimated and adjusted twice daily during the first week and then daily until the experiments were terminated. The contents of the aquarium were gently stirred to ensure that the zooplankton was evenly distributed and then two, 20 ml samples were collected. Formalin was added to each sample and all crustacean zooplankton, including the various copepodite stages but not nauplii, were counted. The mean number of zooplankters per 20 ml was used to estimate the number of zooplankters remaining in each aquarium. The same method was used to estimate the density of zooplankton in the stock solution collected from the pond. The required number of zooplankters were then placed into each aquarium by adding a known volume of the stock solution.

Diet at First Feed

Two hundred larvae were placed into each of three aquaria. From the eighth day after the completion of hatching, zooplankton which passed through a 1000 μm mesh were added daily to provide a density of 1000 zooplankters/L. Ten larvae were sampled daily for five days from each aquarium. The gut of each specimen was removed and dissected with the aid of a microscope and the number, type and length of food items present were recorded.

Zooplankton Density and Delayed Feeding

Same-age larvae from three females that had been induced to spawn were placed together in a trough and from these, 200 larvae were counted into each of 34 aquaria on the eighth day after the completion of hatching. Ten of the aquaria were used to determine the effect of zooplankton density on the survival of Murray cod larvae. Five densities of zooplankton were offered to the larvae in the aquaria from day 1 (10 days post-hatching): 250, 500, 1000, 3000, 5000 zooplankters/L. Each treatment was duplicated. The experiment was terminated on day 24 and the mean survival rate of fry from each treatment determined.

Twenty aquaria were used to determine the effect on survival of delayed feeding at two different densities of zooplankton. Larvae were kept without food until days 5, 8, 11, 13 or 15 and then offered zooplankton at densities of 250 or 3000 zooplankters/L. The remaining four aquaria were used as a control treatment, where larvae were offered zooplankton at the two densities commencing on day 1. Each treatment was duplicated. The experiment was terminated on day 17 and the mean survival rate of larvae from each treatment was determined.

The effect of zooplankton density on larval survival rate in aquaria was analysed using a one-way analysis of variance; the effect of delayed feeding, at the two zooplankton densities, on survival rate was analysed using a two-way analysis of variance. Percentage data were normalized using an arcsin transformation.

Pond Experiment

Ponds 1 and 2 were filled and fertilized 5 and 10 days before stocking, respectively. Pond 1 was fertilized with inorganic fertilizer only, and pond 2 with inorganic fertilizer plus lucerne hay as recommended by Rowland (1989b). Larvae were stocked on day 1 into the ponds at densities of 3 and $19/m^2$. The diets of cod larvae and fry were analysed using the frequency of occurrence and the percent composition by estimated volume methods. The ponds were drained 40 and 34 days after stocking; all fry harvested were counted.

Results

Laboratory Experiments

At water temperatures of 20-22°C, Murray cod eggs generally complete hatching on the eighth day after fertilization and larvae remain clumped together in a single layer on the bottom of troughs for another six or seven days before gradually dispersing; yolk sac absorption is complete nine to 11 days after the completion of hatching (Rowland, 1985, 1988).

Diet at First Feed

Many Murray cod larvae in aquaria commenced feeding on copepodids, copepods and cladocerans (length 220-370 μm) nine and ten days after the completion of hatching; all larvae sampled on the eleventh day contained zooplankton up to 450 μm in length (Table 1). The zooplankton consumed by the larvae were predominantly the copepods, *Boeckella fluvialis* and *Calamoecia lucasi* and the cladocerans, *Moina micrura* and *Daphnia carinata*. Although large numbers of rotifers (*Keratella* spp. and *Brachionus* spp.) were also present in all aquaria, only four of the larvae sampled contained rotifers, suggesting that cod larvae feed selectively on the crustacean zooplankton.

Zooplankton Density and Delayed Feeding

Results of the experiments to determine the effects of zooplankton density and delayed feeding on the survival of Murray cod larvae are presented in Table 2 and Fig. 2.

TABLE 1

Diet of Murray cod larvae held in 70-L aquaria and offered zooplankton collected from ponds and passed through a 1000 μm sieve. 30 larvae were sampled each day

Days after completion of hatching	Larvae with food in gut (%)	Range of food types in number and length (μm) per larva			
		Copepoda[a]	Cladocera[b]	Rotifer	Other
8	0	–	–	–	–
9	37	1-2 (220-290)	–	–	–
10	87	1-4 (220-350)	1-2 (290-370)	–	–
11	100	0-7 (250-370)	1-3 (300-450)	0-3 (80-120)	1[c]
12	90	2-19 (250-450)	0-7 (270-520)	0-2 (100-160)	–

a including copepodids; predominantly *Boeckella fluvialis* and *Calamoecia lucasi*.
b predominantly *Moina micrura* and *Daphnia carinata*.
c *Volvox*; probably ingested unintentionally.

If feeding commenced on day 1, there was no significant difference ($F_{4,5}$ = 1.78, $P>0.05$) between the survival of larvae offered zooplankton at densities of 250-5000 zooplankters/L (Table 2). However a delay in the commencement of feeding resulted in significantly reduced survival ($F_{4,15}$ = 74.02, $P<0.01$), and there was high mortality of cod larvae not fed until day 13. Zooplankton density did have a significant effect (F = 11.98, $P<0.01$) when initial feeding was delayed, and the survival of larvae offered 250 zooplankters/L commencing on days 5, 8, 11 or 13 was lower than the survival of larvae offered 3000 zooplankters/L (Fig. 2).

TABLE 2

Survival of Murray cod larvae offered different densities of zooplankton commencing on the tenth day after the completion of hatching (day 1); experiment terminated on day 24

Density of zooplankton (no./L)	250	500	1000	3000	5000
Mean survival ± S.D. (%)	80.7 ± 7.5	83.0 ± 3.5	88.7 ± 5.3	89.5 ± 3.5	89.9 ± 3.4

From day 7, unfed larvae appeared weaker and less active than larvae that were being fed. By day 12 most unfed larvae were moribund and floated with the gentle current and most of these larvae appeared too weak to capture zooplankton when it was first offered on day 13. Unfed larvae were all dead by day 15.

Diets of Larvae and Fry in Ponds

Summaries of the diets of Murray cod sampled from the ponds are given in Table 3. Larvae sampled from the ponds two days after stocking were feeding predominantly on the copepods *Boeckella fluvialis* and *Calamoecia lucasi* and on the cladoceran *Moina micrura*; these prey items ranged in length from 180 to 450 μm. By day 7, larvae were eating zooplankton, including *Daphnia carinata* up to 2200 μm in length. Chironomid larvae and aquatic insects (mainly notonectids and corixids) were major components of the diet of fry from the third week after stocking until harvest. This shift in diet may have been partly due to the decrease in availability of larger zooplankton in these two ponds.

A total of 42,870 fry were harvested, with survival rates of 62 and 74% in ponds 1 and 2 respectively (Table 4).

TABLE 3

Diet of Murray cod larvae and fry in earthen ponds. Length range of food type in parentheses; zooplankton μm, chironomid larvae and aquatic insects mm

Pond	Days post-stocking	Mean length of 5 larvae/fry (mm)	Frequency of occurrence; percent composition by volume					
			Copepoda	Cladocera	Unidentified zooplankton	Chironomid larvae	Aquatic insects	Unidentified
1	2	13.9	0.8; 64 (180-350)	0.8; 36 (250-450)	—	—	—	—
	7	16.3	1.0; 70 (350-2000)	1.0; 30 (450-2200)	—	—	—	—
	14	20.7	0.8; 40 (500-1800)	0.6; 30 (600-1800)	0.2; 18 —	0.2; 12 (7)	—	—
	21	23.2	0.8; 28 (1800-2900)	0.4; 15 (2200-3200)	0.4; 15 —	0.6; 42 (4-9.5)	—	—
	30	30.1	0.2; 5 (2000)	0.4; 5 (2000-3000)	—	0.8; 60 (3.8-11)	0.4; 20 (4-12)	0.6; 20 —
	40	38.9	—	—	—	0.6; 34 (6-10)	0.6; 46 (9-12)	0.8; 20 —
2	2	13.1	1.0; 70 (220-350)	0.8; 30 (300-450)	—	—	—	—
	7	15.3	0.8; 50 (400-950)	0.4; 10 (550-1400)	0.4; 20 —	0.4; 20 (2.5-5.5)	—	—
	14	20.4	0.8;30 (450-1500)	0.2; 1 (1500)	0.4; 10 —	0.8; 41 (4-9)	0.4; 18 (3-5.5)	—
	21	22.5	0.8; 18 (700-2800)	0.4; 4 (2800-4000)	—	1.0; 60 (4-11)	0.2; 4 (9-12)	0.4; 14 —
	28	26.5	0.6; 18 (650-2100)	0.4; 6 (600-1900)	—	1.0; 46 (4-11.8)	0.6; 20 (10-14)	0.6; 10 —
	34	31.0	0.2; 4 (1200-2100)	0.2; 6 (1800-2500)	—	1.0; 70 (6.5-12.8)	0.4; 10 (11-14)	0.4; 10 —

Fig. 2. Effect of delayed feeding at two densities of zooplankton on the survival of Murray cod larvae. Experiment terminated on day 17. mean survival ± S.D. at zooplankton density of 3000/L; mean survival ± S.D. at zooplankton density of 250/L.

TABLE 4

Details of Murray cod stocked in ponds

Pond	Surface area (m^2)	No. of larvae	Stocking density (no./m^2)	No. of fry harvested	Survival (%)	Stocking period (days)
1	3,000	59,000	19	36,580	62	40
2	2,800	8,500	3	6,290	74	34

Discussion

In contrast to the larvae of some other fishes which require densities of zooplankton of 1000/L or greater for survival under intensive rearing conditions (May, 1974), densities of 250-5000 zooplankters/L did not affect the survival of Murray cod larvae. The larvae of *M. peelii* are relatively large (total length, 12-13 mm) and well developed at the completion of yolk sac absorption, and they can swim strongly and effectively capture food items even at the low density of 250 zooplankters/L. However, if initial feeding is delayed for five or more days, survival is significantly affected by food density and a delay of 13 days in initial feeding results in high mortality irrespective of zooplankton density.

Most fish larvae show a rapid increase in feeding efficiency as they grow (May, 1974), but if sufficient food (of suitable size) is not encountered, swimming ability will decline (Laurence, 1972). This was observed in unfed Murray cod larvae from day 7; by day 12 most unfed larvae were moribund. Because fish larvae do not have lipid reserves, protein metabolism results in degeneration of the pancreas in starved or poorly fed larvae (O'Connell, 1976) and this can lead to a loss of ability to digest food even if it is encountered and eaten (Dabrowski, 1982; Pitcher and Hart, 1982).

The results of this study suggest that the availability of zooplankton within two weeks of the completion of yolk sac absorption is an important factor influencing survival in *M. peelii*. Rising water levels and floods in the Murray-Darling river system are followed by an increase in plankton and aquatic insects, particularly chironomid larvae (Frith, 1959; Shiel, 1980; Maher and Carpenter, 1984) and these are the major food items of Murray cod larvae and fry in earthen ponds at the Inland Fisheries Research Station. Consequently floods, besides increasing the available habitat for fishes, also probably provide optimum feeding conditions for *M. peelii* larvae and fry in the wild.

The construction of dams, high-level weirs and levee banks on the major tributaries of the Murray-Darling river system has altered the natural flow and temperature regimes and dramatically reduced the frequency, extent and duration of flooding (Reynolds, 1976; Cadwallader, 1978; Walker *et al.*, 1978; Walker, 1979). Consequently optimum conditions for the high survival of Murray cod larvae now rarely occur and poor larval recruitment may be a reason for the dramatic decline of *M. peelii* since the 1950's (Rowland, 1989a).

In ponds at the Inland Fisheries Research Station, Murray cod larvae fed selectively on crustacean zooplankton, particularly the calanoid copepods *Boeckella fluvialis* and *Calamoecia lucasi* and the cladocerans *Moina micrura* and *Daphnia carinata*, in preference to the abundant rotifers. The larvae of another large, native, percichthyid the golden perch (*Macquaria ambigua*) also feed selectively on crustacean zooplankton (Arumugam, 1986; Rowland, 1986). Rotifers generally dominate the zooplankton communities of the Darling River and at times, the Murray River; whereas limnetic zooplankters such as calanoid copepods and cladocerans dominate habitats such as billabongs, backwaters and impoundments (Shiel, 1978, 1979, 1980; Geddes, 1984). The apparent selectivity of the larvae of both *M. peelii* and *M. ambigua* for crustacean zooplankton suggests that billabongs and backwaters in the Murray-Darling river system play an important role in the survival of larvae of these native fishes.

In fish culture, the larval rearing phase is often a 'bottle neck' in production, because of the high mortality that is encountered soon after the commencement of exogenous feeding; particularly in species with very small (<5 mm) larvae. In contrast, Murray cod larvae are relatively easy to feed under culture conditions because they are large and well developed at the completion of yolk sac absorption. Within 7 days larvae can consume copepods and cladocerans as long as 2200 μm (Table 3).

Murray cod larvae can be reared in intensive facilities using newly-hatched brine shrimp (*Artemia*) as the only food item (Cadwallader and Gooley, 1985). Rowland (1985) compared the survival and growth of larvae feeding on natural zooplankton in a pond, troughs and aquaria, and found that although survival rates were similar (83%, 93% and 89% respectively), the growth rate was significantly faster ($P < 0.01$) in ponds. It was suggested that lower stocking density, the presence of cover, natural light intensities and the availability of different food types, such as chironomid larvae and aquatic insects for older larvae and fry, were contributing factors. In the current study, Murray cod grew rapidly and reached mean lengths of 31.0 and 38.9 mm in 34 and 40 days respectively (Table 3).

Techniques for the preparation and management of earthen ponds for rearing larvae of some Australian native fishes were recommended by Rowland (1989b). These procedures, plus supplementary feeding with wild zooplankton and/or brine shrimp in intensive facilities for three to five days prior to stocking, are now used to achieve high survival and growth rates of Murray cod at commercial native fish hatcheries (Bruce Malcolm, Ray Mepham, 1990, pers. comm.).

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